

**AMENDMENTS TO THE CLAIMS**

Please amend claims 20-22, and cancel claim 23, as set forth below. Please withdraw claims 1-19 and 26-28, without prejudice or disclaimer.

The current listing of claims replaces all prior listings.

1. (Withdrawn) A C3A clonal cell line derived from a parental C3A cell line, wherein said clonal cell line has a doubling time in serum-free medium significantly less than the doubling time of said parental line in said serum-free medium.
2. (Withdrawn) The clonal cell line of claim 1, wherein the doubling time in serum-free medium of said clonal cell line is less than about 70% of the doubling time in serum-free medium of said parental C3A cell line.
3. (Withdrawn) The clonal cell line of claim 1, wherein said doubling time in serum-free medium of said clonal cell line is in the range of less than about 50% to less than about 70% of the doubling time in serum-free medium of said parental C3A cell line.
4. (Withdrawn) The cell line of claim 1, wherein cells of said cell line cultured in serum-free medium express a single or any combination of a plurality of harvestable polypeptides.
5. (Withdrawn) The cell line of claim 4, wherein said cells express alpha fetal protein (AFP).
6. (Withdrawn) The cell line of claim 4, wherein said cells express human albumin.
7. (Withdrawn) The cell line of claim 4, wherein said cells express  $\alpha$ -1-antichymotrypsin.
8. (Withdrawn) The cell line of claim 4, wherein said cells express  $\alpha$ -1-antitrypsin.

9. (Withdrawn) The cell line of claim 4, wherein said cells express antithrombin III.
10. (Withdrawn) The cell line of claim 4, wherein said cells express complement C3.
11. (Withdrawn) The cell line of claim 4, where said cells express Factor V.
12. (Withdrawn) The cell line of claim 4, wherein said cells express transferrin.
13. (Withdrawn) The cell line of claim 4, wherein said cells express a single or any combination of a plurality of harvestable polypeptides selected from the group consisting of: alpha fetal protein (AFP), human albumin,  $\alpha$ -1-antichymotrypsin,  $\alpha$ -1-antitrypsin, antithrombin III, complement C3, Factor V and transferrin.
14. (Withdrawn) The cell line of claim 1, said cell line having an ATTC accession No. of CRL-12461.
15. (Withdrawn) A method of producing a single or any combination of a plurality of harvestable polypeptides, comprising:
  - a) culturing cells of the cell line of claim 1 in serum-free medium,
  - b) expressing said polypeptide/s from said cells; and
  - c) recovering said polypeptide/s from said culture to produce a harvestable polypeptide.
16. (Withdrawn) A method of producing the cell line of claim 1, comprising:
  - a) sequentially culturing cells of a parental C3A cell line in a series of medium having incrementally decreasing concentration of serum, the final medium in said series being serum free,
  - b) generating a clonal cell colony of said cells from said final medium in said series of
    - a) in serum-free medium; and
    - c) propagating said colony in serum-free medium to produce a serum-free cell line.

17. (Withdrawn) The method of claim 16, wherein one of said series of medium having incrementally decreasing concentration of serum in said sequential cultures series has a ratio of serum containing and serum-free medium of about 50:50.

18. (Withdrawn) The method of claim 16, wherein one of said series of medium having incrementally decreasing concentration of serum in said sequential cultures series has a ratio of serum containing and serum-free medium of about 25:75.

19. (Withdrawn) The method of claim 16, wherein the serum-free medium is JRH Bioscience ExCell 620 supplemented with 2mM L-glutamine.

20. (Currently Amended) An extracorporeal bio-artificial liver device comprising an apparatus containing cells of the cell line deposited as ATCC accession No. CRL-12461, wherein the cells have a doubling time in serum-free medium which is less than about 70% of the doubling time in serum-free medium for C3A cells ~~or cells clonally derived from cells deposited as ATCC accession No. CRL-12461~~, wherein the ~~clonally derived~~ cells are cultured in serum-free medium on a surface in the device in an amount and having liver specific biological activity at a level sufficient to sustain a subject having a liver disorder or compromised liver function, and wherein the surface is contained within a hollow fiber cartridge, wherein the hollow fiber is formed from a material which has a pore size of about 0.1  $\mu$ m to 0.3  $\mu$ m, and wherein the cartridge is at least 1400 cm<sup>2</sup>.

21. (Currently Amended) A method of using cells of the cell line deposited as ATCC accession No. CRL-12461, wherein the cells have a doubling time in serum-free medium which is less than about 70% of the doubling time in serum-free medium for C3A cells ~~or cells clonally derived from cells deposited as ATCC accession No. CRL-12461~~ in an extracorporeal bio-artificial liver device, comprising:

a) providing the cells to a surface in the device, wherein the device comprises a hollow fiber cartridge formed from a material which has a pore size of about 0.1  $\mu\text{m}$  to 0.3  $\mu\text{m}$ , and wherein the cartridge is at least 1400  $\text{cm}^2$ ;

b) culturing the cells in the device in serum-free medium;

c) attaching the extracorporeal device to a subject between an artery and vein of the subject, wherein the device is in fluid communication with the artery and vein;  
and

d) perfusing blood from the subject through the attached device,

wherein the cultured cells interact with the blood to provide bio-artificial liver support for the subject.

22. (Currently Amended) A method of treating a subject having compromised liver function, comprising:

a) providing cells of the cell line deposited as ATCC accession No. CRL-12461, wherein the cells have a doubling time in serum-free medium which is less than about 70% of the doubling time in serum-free medium for C3A cells ~~or cells clonally derived from cells deposited as ATCC accession No. CRL-12461~~ to a surface in an extracorporeal bio-artificial liver device, wherein the cells are provided in an amount and having liver specific biological activity at a level sufficient to sustain said subject having said compromised liver function,

b) culturing said cells in said device in serum-free medium, , wherein the device comprises a hollow fiber cartridge formed from a material which has a pore size of about 0.1  $\mu\text{m}$  to 0.3  $\mu\text{m}$ , and wherein the cartridge is at least 1400  $\text{cm}^2$ ;

and

c) perfusing blood from said subject through the device to contact said cells,  
wherein the perfusing results in ~~removal of blood borne toxic solutes entering said device~~ and release of protein and low molecular weight products from said cells into blood exiting said device.

23. (Canceled)

24. (Previously Presented) The method of claim 22, wherein the compromised liver function is associated with Fulminant hepatic failure (FHF).
25. (Original) The method of claim 22, wherein said subject is a human.
26. (Withdrawn) A method of producing protein comprising:  
a) culturing cells of any of claims 1 to 14 in serum-free medium to express a single or any combination of a plurality of harvestable polypeptides; and  
b) recovering said polypeptide/s to produce protein.
27. (Withdrawn) A method of screening compounds for metabolic activity comprising:  
a) providing a compound to cells of the cell line of claim 1, wherein said cells are cultured in serum-free medium; and  
b) analyzing said cells for the presence of metabolites of said compound to screen for metabolic activity.
28. (Withdrawn) A method of studying enteric disease comprising:  
a) providing a bacterial organism to cells of the cell line of claim 1, wherein said cells are cultured in serum free medium; and  
b) employing said cells of a) for experimental use to study enteric disease.
29. (Previously Presented) The method of claim 24, wherein the protein is albumin.